Oil Immersion Condensers

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One of the most important equations in optics is the law of refraction, often called Snell's law.

$$\frac{\sin\theta_1}{\sin\theta_2} = \frac{v_1}{v_2} = \frac{n_2}{n_1}$$

The θ's are the angles from the normal to the light path. The v's are the velocities of light, and the n's are the indices of refraction.

We can also write Snell's law thus:

$$\sin\theta_i = \frac{n_2}{n_1}\sin\theta_r$$

Where θ_i is the angle of incidence, and where θ_r is the angle after refraction. When $\sin\theta_i$ is equal to 1.0 we have the maximum angle of incidence, often called the critical angle. Thus

$$\theta_c = \arcsin\left(\frac{n_2}{n_1}\right)$$

In microscopy numerical aperture, often abbreviated "na", is defined as $\mathbf{na} = n\sin\theta$. (θ is the angular radius of the cone of light that is taken in by the lens. n is the index of refraction of the medium between the lens and the object.)

The angular aperture is the angle across the cone of light taken in by an objective. It is commonly denoted by α. This is twice the angular radius of the cone of light. Thus $\theta = \frac{\alpha}{2}$

For water at 20C n is 1.3330, and for the glasses used for slides, those used for most "outer" microscope glass lens surfaces, and standard microscope immersion oils it is 1.515.

Thus the critical angle from water to air is 48.607^o.

The critical angle from immersion oil or glass to air is 41.305^o.

The critical angle from immersion oil or glass to water is 61.626^o.

Many oil immersion dark field condensers create a hollow cone inside the top of condenser, oil, and glass in contact with the oil having a numerical aperture of 1.4 with the field stop covering everything below 1.2.

The angle in oil at 1.4 na is is 67.53^o. ($\arcsin\frac{1.4}{1.515}$)

The angle in oil at inner edge with 1.2 na is 52.38^o ($\arcsin\frac{1.2}{1.515}$)

Note that the critical angle from glass to water is 61.626^o. The outer part of the cone is beyond the critical angle of this interface! This part of the condenser's light undergoes total internal reflexion at the glass-water interface. The remaining light continues on until it either interacts with the specimen or reaches the cover slip. Should it get to the over slip it continues on until it hits the glass air surface, and there all the light that has not interacted with the specimen undergoes total internal reflexion back down through the system.

Oil immersion condensers are designed to absorb light that is internally reflected back into them, unless it emerge the second time into the 1.2 to 1.4 na part of the cone.

Should the specimens be in actual contact with the upper surface of the slide, they can scatter or refract light light that is beyond the critical angle. Because of total internal reflection, there may be two chances for specimen objects to refract or scatter light.

Oil immersion dark field condensers have a tremendous advantage over dry dark field condensers because the light that passes directly through the condenser, slide, specimen, and cover glass never can emerge from the cover slip. Thus dry objectives up to and including na=0.95 apochromats can be used with them! One can get decent resolution with dark field this way!

The shallow angle of the light beam relative to the object plane, however, means that a relatively small area of the specimen is illuminated. Dark field condensers are thus generally not appropriate for low power objectives.

There is another way that oil dark field condensers can be used to produce high resolution images. Many manufacturers manufacture special oil immersion objectives at 90x to 100x with iris diaphragms.

One can adjust the iris until the na is just small enough to produce a dark field. Remember with an oil immersion objective, the light passes unrefracted from the condenser into the objective. Thus an iris is needed! Some also make lower power water immersion objectives this way.

Here are some hints about using oil immersion condensers:

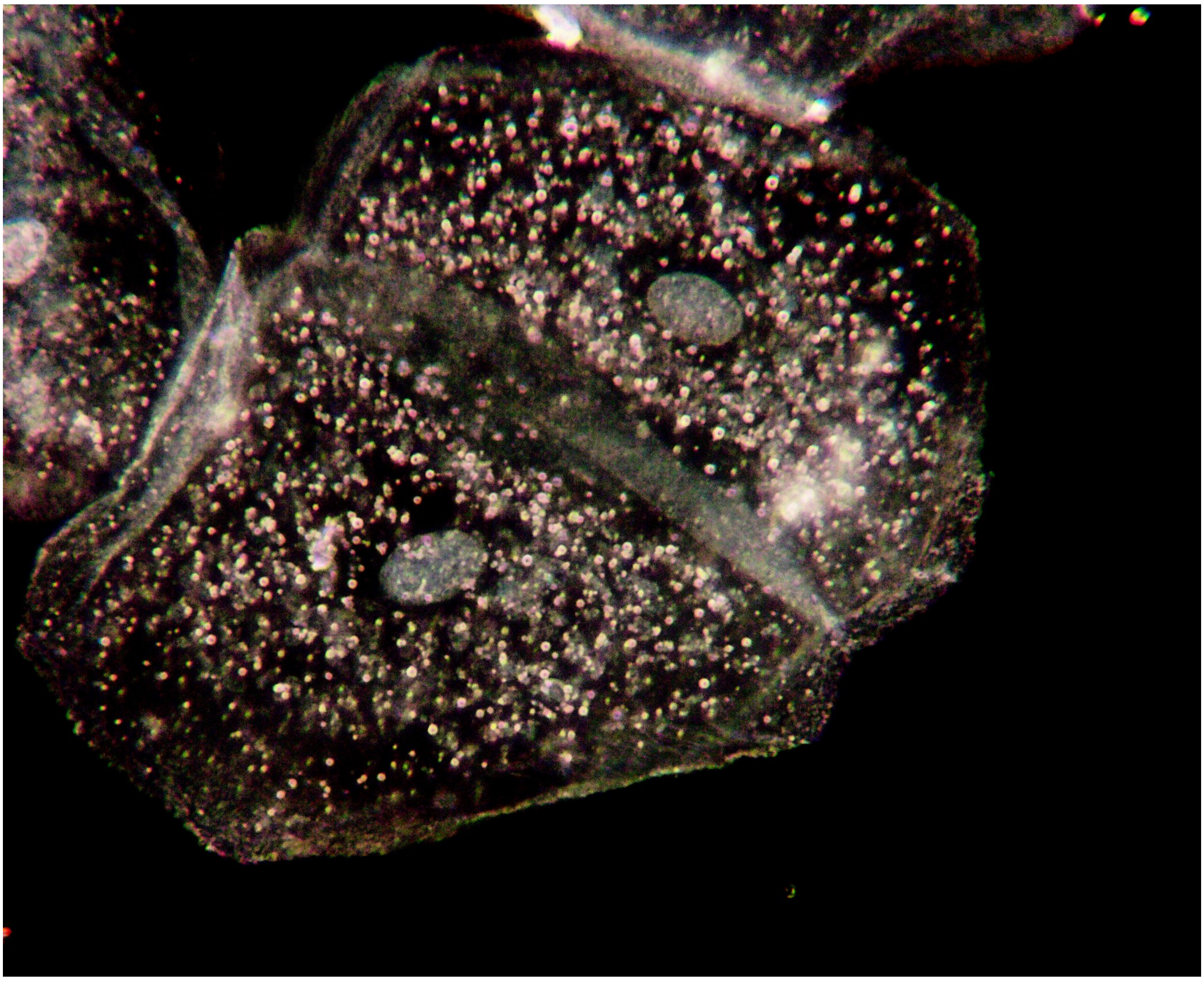
- Place the immersion oil in the centre of the upper surface of the condenser oil with the condenser a bit below the point where the slide will be. Do not put too much, or it may run down the sides of the condenser. Do not use high viscosity immersion oil because it will tend to have bubbles.
- After the oil is on top of the condenser, place the slide on the stage and turn on the illuminator. Move the condenser focus toward the base of the slide until full contact is made. Take care not to push the slide above the plane of the stage.
- Most people find it best to centre the condenser using a low power objective. Oil immersion condensers illuminate a relatively small disk on the object plane. Once the condenser is centred at low power, one can go to higher powers. If using an oil immersion lens with an iris, it is best to be certain the condenser is centred with a dry 40x or 60x before using the oil immersion lens.
- Oil immersion condensers coupled with high na high power objectives permit extremely small objects on the slide to be visible. Small things will appear as dots, they can be far smaller than the wavelength of light being used for illumination. However, such dots will appear dimmer and dimmer for smaller and smaller objects. Because very small objects are visible, slides and cover slips need to be very clean!
- Small out of focus objects will appear as rings. When the rings are incomplete it is a sign that there is not enough oil between the surface of the condenser and the under side of the slide.
- From what has been said above, high power high resolution dark field observation requires either very high na dry objectives or immersion objectives. These types of objectives tend to be expensive, especially high na dry ones. Procedures are simpler with high na dry objectives, but immersion iris objectives may make it possible to get a bit higher resolution, but at the expense of simplicity! In the past it was often possible to find several different kinds of water and oil immersion iris objectives that were produced by LOMO and sold for fairly low prices. LOMO also produced a number of 1.0 na water immersion lenses. LOMO objectives seem to be increasingly difficult to find.
- Almost all oil immersion objectives used at full aperture will take in the light from oil immersion condenser. It is possible to obtain (or make) a so called "funnel stop" to insert into the back of the objective to "stop it down".

Sample test image:

Epithelial cells have become almost traditional as a test subject. The image on the next page is taken using a Tiyodo dark field na=1.2-1.4 oil immersion condenser on an 1969 Orthoplan microscope. The objective used is a 40x Leitz na=0.95 apochromat. A Toupcam 4096x3288 microscope camera was used to obtain the image. This camera is designed to utilise standard C camera fittings or else special reducing adaptors to connect to standard microscope ocular tubes. Here it was attached to the standard Orthoplan C adapter placed on the photo port of the standard 80-10-10 Orthoplan head. (The reducing adapters were not used.)

This particular oil immersion dark field condenser has a special toroidal lens beneath the condenser to increase the amount of light getting into the optical system. This dramatically increases the brightness of the image. (This particular type of oil immersion condenser seems extremely difficult to locate. This particular example has an Leitz dove tail mount.)

Notice how tiny out of focus objects in the cell appear as tiny rings. The depth of field for a 40x 0.95 na objective is very shallow!



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